

ORIGINAL RESEARCH

Intradermal positivity after negative prick testing for inhalants

Shawn P. McKay, MD, Danny Meslemani, MD, Robert J. Stachler, MD, and John H. Krouse, MD, PhD, Detroit, Michigan

OBJECTIVE: Examine the incidence of positive intradermal tests after a negative skin prick test for 24 inhalant antigens.

STUDY DESIGN AND SETTING: Retrospective study. Charts from patients who underwent modified quantitative testing (MQT) over a 3-year period were reviewed. Patients were initially tested with Multi-Test II. Subjects with negative wheals to a specific allergen were then tested with a 1:500 weight:volume intradermal injection of that allergen.

RESULTS: One hundred thirty-three patients underwent MQT for 24 antigens. Allergens with the highest incidence of positive intradermal wheals after negative prick testing were *Dermatophagoides pteronyssinus* and *Dermatophagoides farinae* at 26.67%. Allergens with elevated incidence of positive intradermals included fusarium, cockroach, cocklebur, rough marsh elder, and ragweed, all with incidences of 16% to 19%.

CONCLUSION: Positive intradermal responses after negative prick testing occur commonly. Future study is needed to determine the generalizability and clinical significance of these findings.

EBM rating: C-4

© 2006 American Academy of Otolaryngology–Head and Neck Surgery Foundation. All rights reserved.

Among the challenges facing the allergist is deciding which testing techniques should be routinely used to diagnose clinically relevant allergies. Allergy testing would ideally demonstrate a high degree of sensitivity and specificity, and could be performed quickly, safely, and with relatively little cost. It would also provide quantifiable information regarding sensitivity of individual antigens that can be used to direct successful treatment.

The type of testing most widely used for evaluating inhalant allergy is skin prick testing (SPT). An advantage of

SPT is that it can be performed quickly; it also utilizes fewer resources than other forms of skin testing.¹ SPT may be used as a stand-alone technique, or it may be used in the context of other testing methods.

Another variation of allergy skin testing is intradermal testing. One form of this technique used by many otolaryngologists is intradermal dilutional testing (IDT). This technique is based on skin endpoint titration (SET), a specific form of IDT in which 1:5 serial dilutions are used progressively to quantify the degree of sensitivity to specific antigens. SET was formalized by Rinkel² and has provided a common nomenclature that other forms of IDT have drawn upon. A drawback to SET is that it requires a substantial amount of time to perform, utilizes significant resources, and may be accompanied by more patient discomfort than other testing techniques.¹

A variation of IDT that has been developed over the past several years is modified quantitative testing (MQT) (Fig 1). This technique initially uses skin prick testing to determine an approximate range of sensitivity, followed by a single intradermal test to further identify the level of sensitivity and quantify the allergic response.³ MQT benefits from the speed and ease of skin prick testing, yet it also provides quantification of sensitivity, which is the primary benefit of other forms of IDT. It has also been shown to correspond very well quantitatively to SET.⁴

One question that has arisen in the literature is the significance of positive intradermal tests after negative SPT. The MQT technique is to place a #2 intradermal (1:500 w:v) when there is a negative skin prick test (Fig 1). The rationale behind this approach is that an SPT with Multi-Test II has

From the Department of Otolaryngology–Head and Neck Surgery, Wayne State University.

Dr Krouse is a consultant, advisor, and shareholder for Schering Plough and Alcon; a consultant and advisor for Sanofi-Aventis; and a consultant for ALK-Abello. Dr Stachler is a consultant for Sanofi-Aventis.

Reprint requests: John H. Krouse, MD, PhD, Wayne State University, 5E UHC, 4201 St. Antoine, Detroit, MI 48202.

E-mail address: jkrouse@med.wayne.edu.

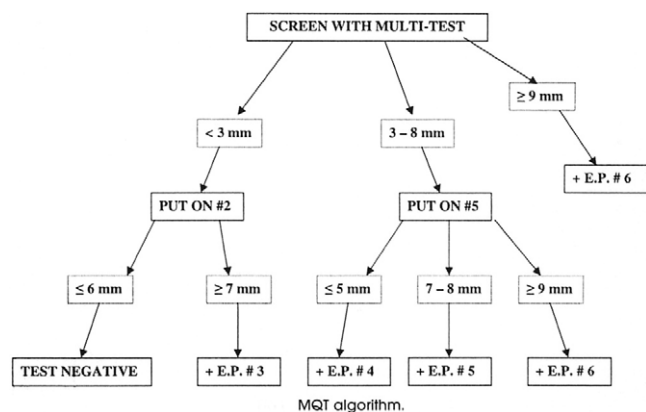


Figure 1 MQT algorithm.

been shown to demonstrate a degree of reactivity relatively equivalent to a #3 intradermal test; a negative prick test, therefore, may overlook less sensitive yet clinically relevant allergy.⁵ When a #2 intradermal is negative after a negative prick, the allergist can be relatively confident that the patient is not sensitive to that antigen. However, when this #2 intradermal test is positive, there is controversy as to the meaning of this result. Several authors have argued that positive intradermals after negative SPT are not helpful in distinguishing clinically significant symptom-producing allergies.⁶⁻⁹

In attempting to answer this question, it is prudent to look at individual antigens and antigen classes. It is possible that the responsiveness may vary for different antigens. Recent work at our institution using nasal provocation testing as a gold standard demonstrated that SPT with Multi-Test II has a high degree of sensitivity (87%) and specificity (86%) when evaluating timothy grass, and that an intradermal increases sensitivity only a minor amount (to 93%).¹⁰ When the same study was performed with *Alternaria* antigen, however, it was discovered that neither SPT nor intradermal testing was especially sensitive or specific in detecting allergy,¹¹ although intradermal testing did add modestly to the sensitivity of the test. These findings suggest that two antigens from different antigen classes may behave differently during skin testing, even though they both can stimulate an effect with provocation at the end organ.

The purpose of this study, therefore, was to retrospectively examine our experience with MQT and to evaluate the frequency with which antigens demonstrate a positive intradermal response after negative prick testing. Based on previous studies from our institution,^{10,11} we hypothesize that there will be variability in intradermal responsiveness, and that mold antigens will be most likely to demonstrate positive intradermal whealing after a negative prick test.

METHOD

This study is a retrospective chart review of patients who underwent MQT between March 2002 and March 2005. The

study was conducted through the offices and clinics of University Otolaryngology, P.C. and the Wayne State University Department of Otolaryngology–Head and Neck Surgery. The protocol was reviewed and approved by the Human Investigation Committee at Wayne State University. Patients who presented to the office and who displayed signs and symptoms of allergic rhinitis underwent allergy skin testing by standard MQT protocol. These patients were tested for 24 antigens, as well as histamine and glycerin controls (Table 1). Patients were initially tested using Multi-Test II (Lincoln Diagnostics, Decatur, IL) as described in the MQT algorithm (Fig 1).³ Those patients who had a reaction of less than 3 mm in 20 minutes for a specific antigen then had a #2 intradermal (1/500 w:v) placed according to the protocol. Patients were considered to have positive intradermal reaction if they produced a wheal of at least 7 mm in diameter. For each patient, all negative prick responses for each antigen were recorded, as well as the number of positive #2 intradermal after the negative prick.

RESULTS

A review of office charts found that 133 patients had undergone MQT during the 3-year reference period. The total

Table 1
Frequency of negative prick testing to 24 antigens (N = 133)

Antigens tested	# negative prick tests	% negative prick tests
White ash	94	70.7
Eastern cotton	107	80.0
American elm	104	78.2
Sugar maple	92	69.2
White oak	97	72.9
American sycamore	100	75.2
Kentucky blue grass/Johnson*	85	63.9
Timothy grass	81	60.9
<i>Alternaria</i>	108	81.2
<i>Aspergillus</i>	118	88.7
<i>Fusarium</i>	123	92.5
<i>Helminthosporium</i>	117	88.0
<i>Hormodendrum</i>	120	90.2
<i>Mucor racemosus</i>	124	93.2
<i>Penicillium</i>	124	93.2
<i>Pullaria</i>	122	91.8
<i>Dermatophagoides farinae</i>	90	67.7
<i>Dermatophagoides pteronyssinus</i>	90	67.7
Cockroach	90	67.7
Cat	79	59.4
Dog	110	82.7
Cocklebur	100	75.2
Rough marsh	100	75.2
Ragweed	86	64.7

*Test with either Kentucky blue grass or Johnson grass.

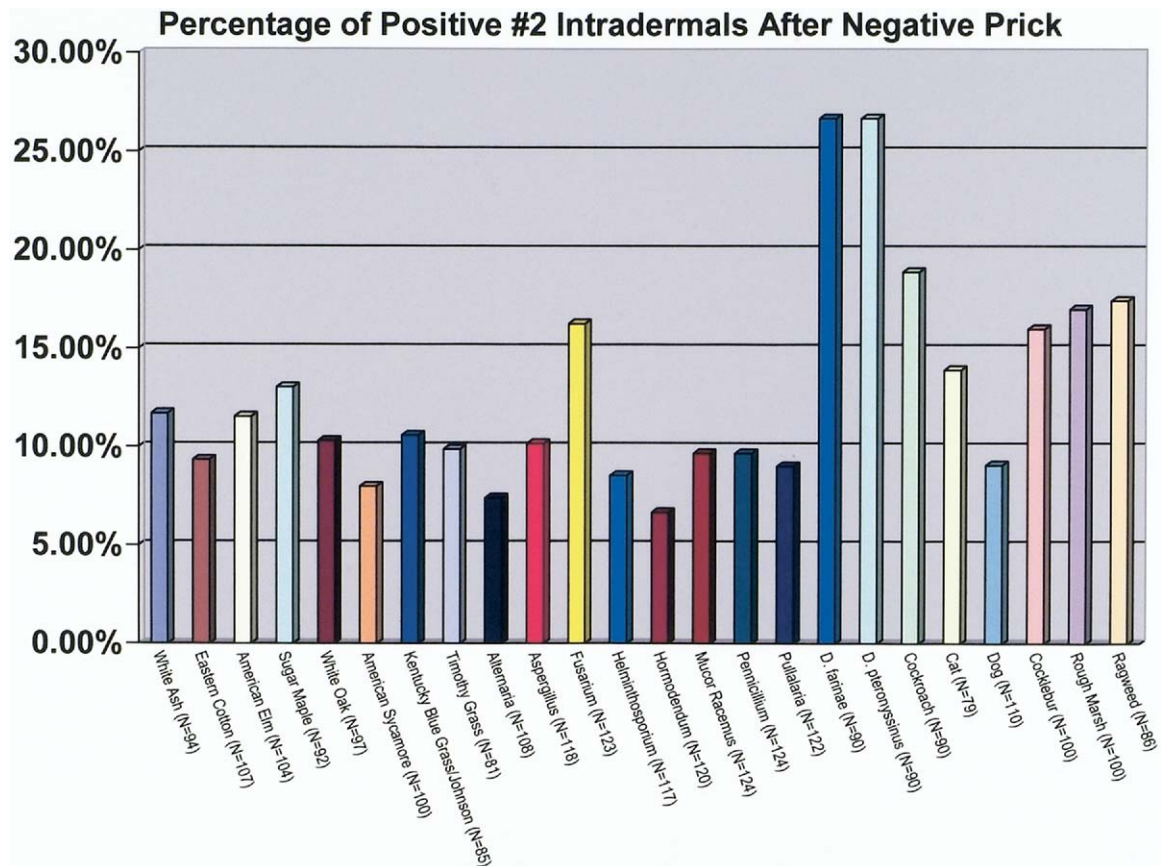


Figure 2 Percentage of negative pricks associated with positive #2 intradermal whealing.

number of negative pricks per antigen for these 133 patients is displayed in Table 1. The percentage of these negative pricks that was associated with positive #2 intradermal whealing is displayed in Figure 2. Allergens with the highest incidence of positive #2 intradermal tests (>7 mm) after negative prick testing were the dust mites *Dermatophagoides pteronyssinus* and *Dermatophagoides farinae*, both with 26.67%. Allergens with elevated incidence of positive intradermals included fusarium, cockroach, cocklebur, rough marsh elder, and ragweed, all with incidences of 16% to 19%.

DISCUSSION

Otolaryngic allergists have historically practiced the classic form of IDT formalized by Rinkel,² known as skin endpoint titration (SET). SET has afforded practitioners the benefit of demonstrating the presence or absence of allergy, as well as quantifying the degree of sensitivity for each antigen. This method has allowed practitioners to find a safe yet potent starting dose for desensitization immunotherapy.³ SET has also provided a common practice approach and vocabulary that other more recent forms of IDT have been able to draw upon, most importantly its use of 1:5 serial dilutions and common nomenclature.

One variation of IDT that is commonly employed is modified quantitative testing (MQT) (Fig 1). This technique blends epicutaneous skin prick testing with intradermal testing and approximates a quantitative endpoint based on SET. One question that has arisen in the literature is whether it is helpful to perform an intradermal test after a negative skin prick test. Traditionally, intradermal testing has been thought to be more sensitive than SPT and has therefore been used when SPT is negative. Also, since Multi-Test II approximates the quantitative reactivity of a #3 intradermal, it has been thought that relying on this device alone may cause practitioners to miss less sensitive yet clinically relevant allergies. However, it is not clear whether a reaction to a #2 intradermal represents a true positive response or rather a nonspecific local skin reaction. It has been noted, for example, that glycerin or lysine-sugar groups may cause local responses via irritant or other non-IgE-mediated mechanisms.¹²

Schwindt et al recently examined patients who had positive intradermals after negative SPT and reported that even when positive intradermal testing did correlate with nasal challenge results, these findings did not necessarily correspond with patient history or symptoms. Among 14 patients in this study with negative SPT and positive intradermals, 6 did not have reproducible intradermals, and 2 did not have a history of allergic rhinitis.⁶

Gungor et al compared SPT to formal SET using nasal provocation testing as a gold standard and found that information obtained by SPT with Multi-Test is comparable to SET method of IDT in terms of sensitivity, specificity, and overall performance.⁷ In fact, Multi-Test results appeared to correlate slightly better with nasal provocation than did SET testing. This study examined only the single antigen, ragweed.

Nelson et al looked at the role of intradermal testing following negative SPT with timothy grass and concluded that positive intradermal wheals did not indicate the presence of clinically significant allergy among a small sample of 9 patients.⁸ Wood et al also performed intradermal skin tests on patients who had negative SPT for cat allergy.⁹ This study found that intradermal testing may have some value in ruling out clinically relevant cat sensitivity, but that overall the technique appeared to add little to the patient's diagnostic evaluation. The major problem with both of these studies is that they injected 0.02 mL of antigen and used an arbitrary 6-mm wheal as a criterion for positivity rather than the 7-mm diameter employed with IDT and MQT. This technique is different than that historically used by otolaryngic allergists and may lead to false-positive results for intradermal testing.¹²

One problem encountered when evaluating the use of intradermal testing after negative prick is that many studies employ only one or few antigens. Recent findings suggest that not all antigens respond similarly with skin testing but rather can vary in the robustness with which they react to testing. Research from our institution suggests that while skin testing techniques such as Multi-Test II and intradermal testing may be accurate and sensitive methodologies for the diagnosis of allergy to pollens such as timothy grass, the same may not be true for mold antigens such as *Alternaria*.^{10,11} A possible explanation for this finding is that some individual antigens may provoke clinically relevant inflammation that is not mediated through a simple IgE type I hypersensitivity alone. The evaluation of delayed, type IV reactions with intradermal tests, as has been clinically observed with dust mites and molds, may improve sensitivity of testing by identifying non-IgE-mediated hypersensitivity or relevant late-phase reactions.

We performed this study in order to gain a sense where future efforts need to be directed. When attempting to answer the question of the utility of intradermal testing after negative responses to prick testing, our present results imply that allergists may need to examine their testing methods for specific antigens or classes of antigens on an individual basis. We found in our sample that dust mites were the most likely antigens to demonstrate a positive intradermal response after a negative prick, followed in frequency by cockroach, ragweed, rough marsh elder, fusarium, and cocklebur. Simons reported similar findings, noting that dust mites, cockroach, and ragweed are the most likely antigens to demonstrate positive intradermal wheals following negative skin prick testing.¹³ Future studies with end-organ provocation tests, symptom scales, and quality-of-life indices are needed to determine if these individual antigen

results indicate clinically relevant allergy or only false-positive findings. In addition, research is necessary to examine if current testing methods, which primarily evaluate type I IgE-mediated hypersensitivity, are sensitive and specific enough to demonstrate clinically relevant antigen-specific inflammation stimulated through other immune pathways.

CONCLUSION

Recent debate in the literature has examined the importance and interpretation of positive intradermal test results after negative SPT. The current study demonstrates that these positive results occur most commonly with dust mites, but can also be seen with other antigens, particularly fusarium, cockroach, cocklebur, rough marsh elder, and ragweed. Future study is needed to determine the generalizability and clinical significance of these findings.

REFERENCES

1. Shah SB, Emanuel IA. Cost analysis of employing multi-test allergy screening to guide serial endpoint titration (SET) testing versus SET alone. *Otolaryngol Head Neck Surg* 2003;129:1–4.
2. Rinkel HJ. The management of clinical allergy: III inhalant allergy therapy. *Arch Otolaryngol* 1963;77:205–25.
3. Krouse JH, Mabry RL. Skin testing for inhalant allergy 2003: current strategies. *Otolaryngol Head Neck Surg* 2003;129:S33–49.
4. Peltier J, Ryan MW. Comparison of intradermal dilutional testing with the Multi-Test II applicator in testing for mold allergy. *Otolaryngol Head Neck Surg*. *Otolaryngol Head Neck Surg* 2006;134:240–4.
5. Murphree JT, Kniker WT. Correlation of immediate skin test responses to antigens introduced by multi-test and intracutaneous routes. *Ann Allergy* 1979;43:279–85.
6. Schwindt CD, Hutcheson PS, Leu SY, et al. Role of intradermal skin tests in the evaluation of clinically relevant respiratory allergy assessed using patient history and nasal challenges. *Ann Allergy Asthma Immunol* 2005;94:627–33.
7. Gungor A, Houser SM, Aquino BF, et al. A comparison of skin endpoint titration and skin-prick testing in the diagnosis of allergic rhinitis. *Ear Nose Throat J* 2004;83:54–60.
8. Nelson HS, Oppenheimer J, Buchmeier A, et al. An assessment of the role of intradermal skin testing in the diagnosis of clinically relevant allergy to timothy grass. *J Allergy Clin Immunol* 1996;97:1193–1201.
9. Wood RA, Phipatanakul W, Hamilton RG, et al. A comparison of skin prick tests, intradermal skin tests, and RASTs in the diagnosis of cat allergy. *J Allergy Clin Immunol* 1999;103:773–9.
10. Krouse JH, Sadrazodi K, Kerswill K. Sensitivity and specificity of prick and intradermal testing in predicting response to nasal provocation with timothy grass antigen. *Otolaryngol Head Neck Surg* 2004;131:215–9.
11. Krouse JH, Shah AG, Kerswill K. Skin testing in predicting response to nasal provocation with *alternaria*. *Laryngoscope* 2004;114:1389–93.
12. Hurst DS, Gordon BR, Krouse JH. The importance of glycerin-containing negative control tests in allergy research studies that use intradermal skin tests. *Otolaryngol Head Neck Surg* 2002;127:177–81.
13. Simons JP, Rubinstein EN, Kogut VJ, et al. Comparison of Multi-Test II skin prick testing to intradermal dilutional testing. *Otolaryngol Head Neck Surg* 2004;130:536–44.